# Evaluating High Throughput Toxicokinetics and Toxicodynamics for IVIVE

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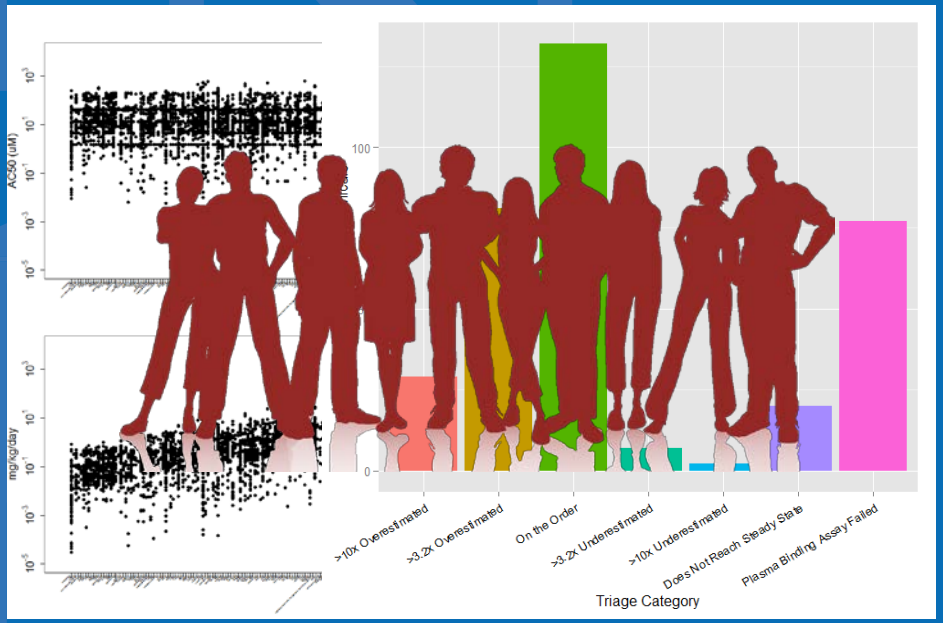


*Figure includes image from Thinkstock*

***IVIVE Approaches***
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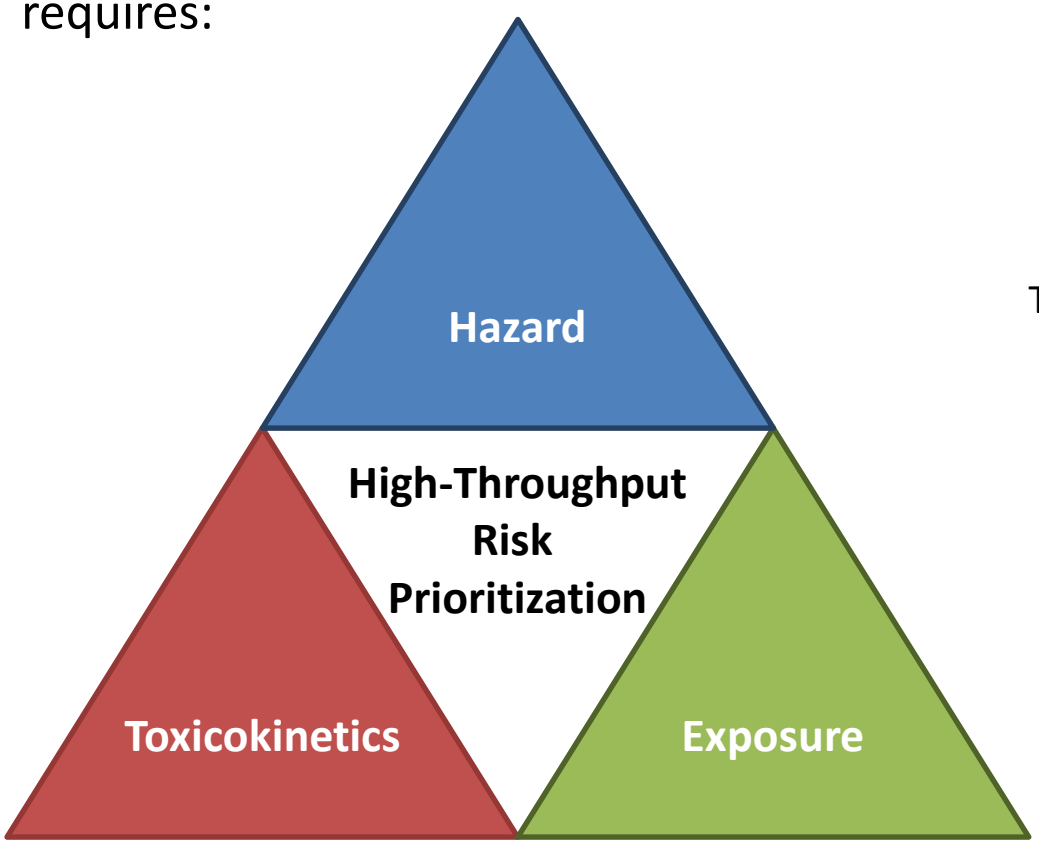
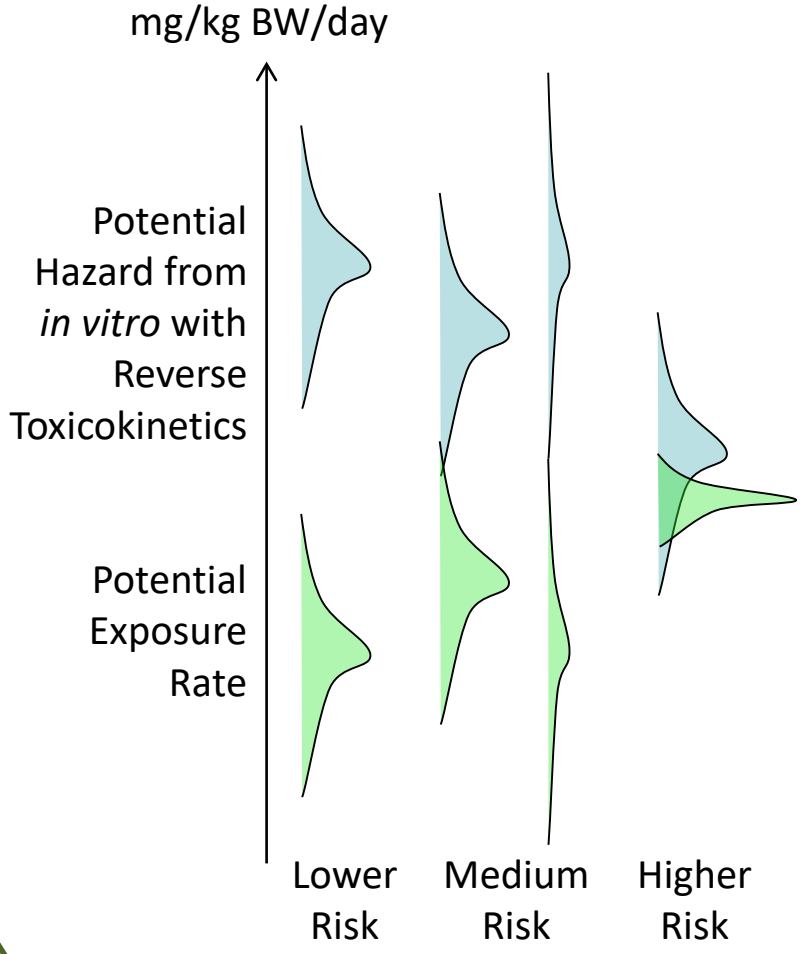
# Introduction

- High throughput risk prioritization based upon *in vitro-in vivo* extrapolation (IVIVE) requires:



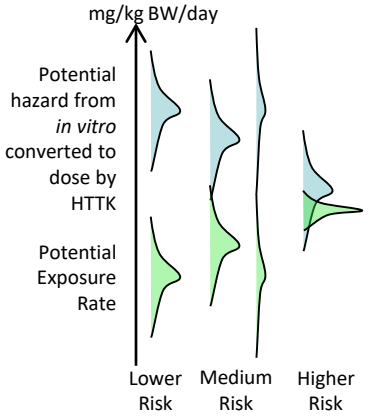




**Most chemicals do not have TK data** – Wetmore et al. (2012…) use *in vitro* methods adapted from pharma to fill gaps

# Toxicokinetic IVIVE:
## Convert HTS µM to mg/kg/day

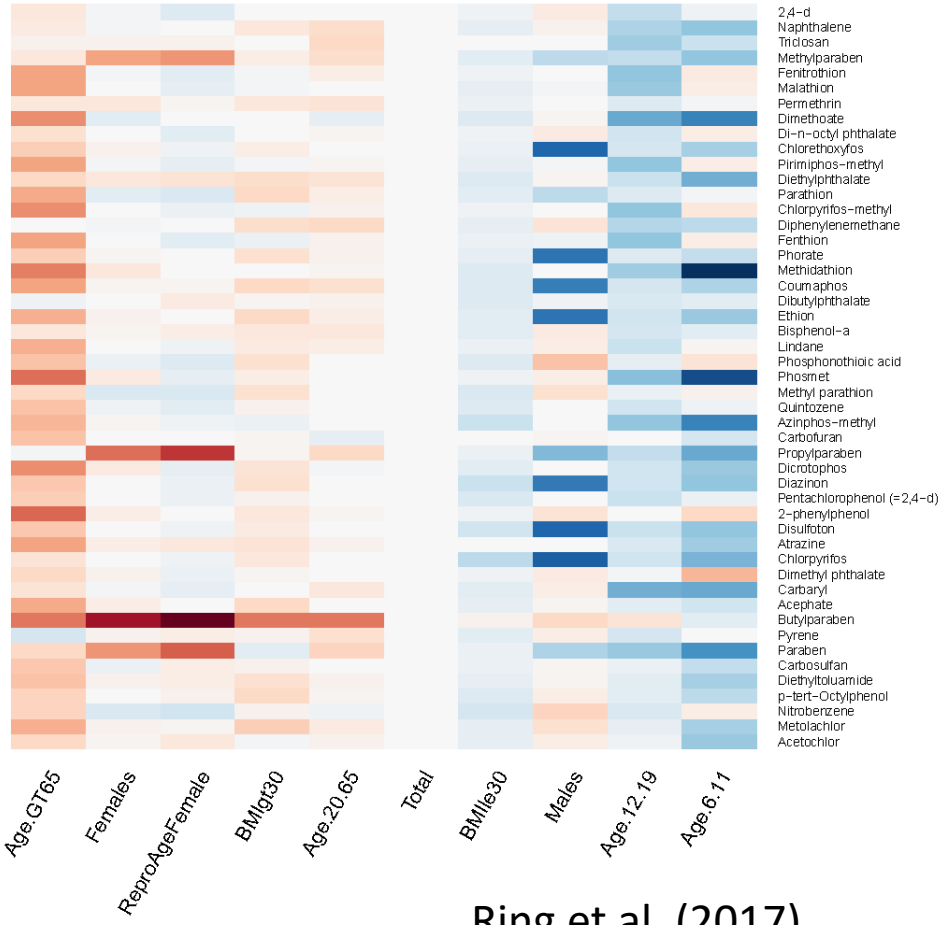
- Can use HTTK to calculate margin between bioactivity and exposure for specific populations

- Using National Health and Nutrition Examination Survey (NHANES) to simulate TK variability and characterize exposure for modern populations




Change in Risk

**Change in Activity : Exposure Ratio**




Ring et al. (2017)

# *In vivo* Predictive Ability and Domain of Applicability

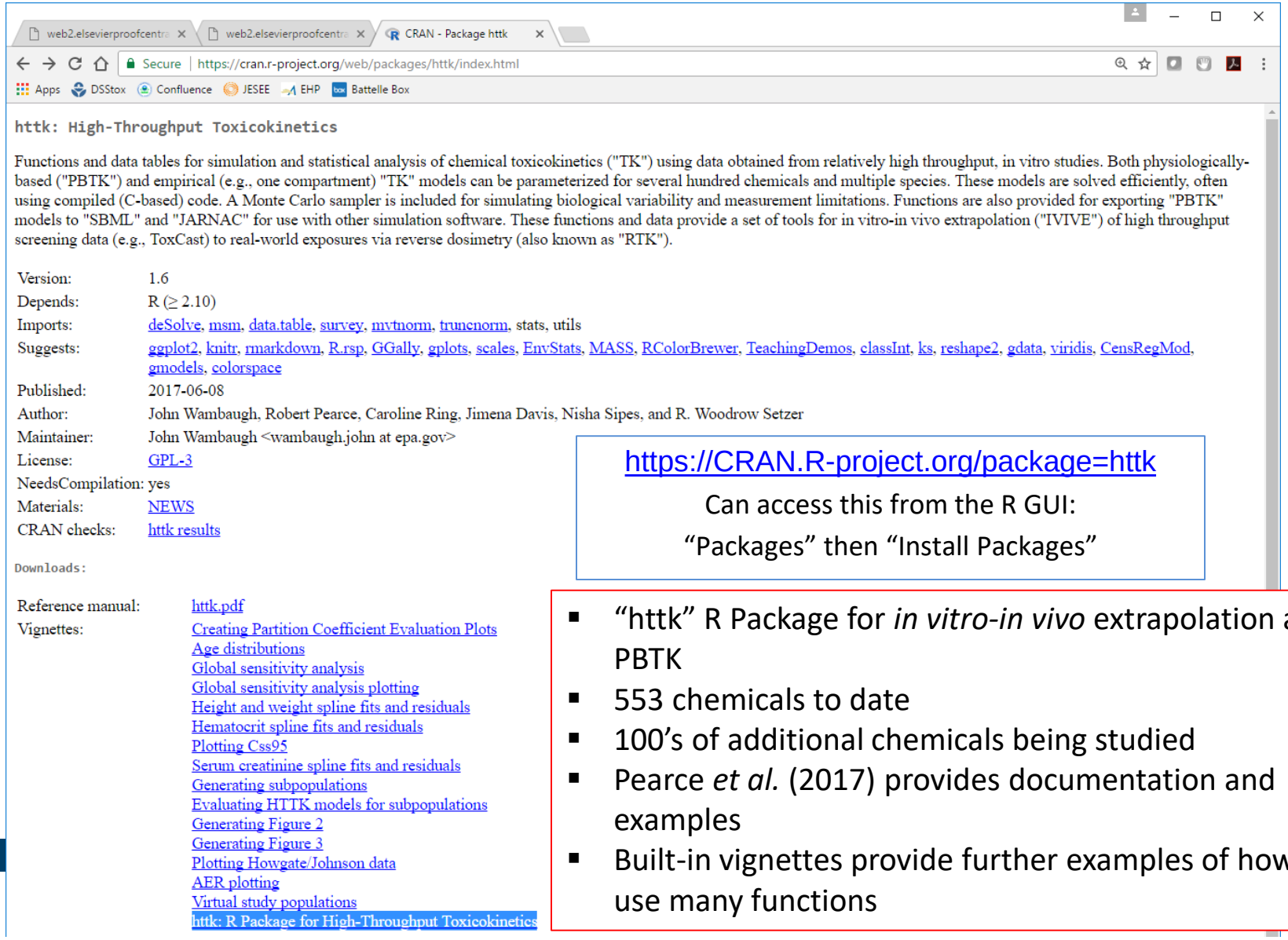
- In drug development, HTTK methods estimate therapeutic doses for clinical studies – predicted concentrations are typically on the order of values measured in clinical trials (Wang, 2010)

- For environmental compounds, there will be no clinical trials

- Uncertainty must be well characterized ideally with rigorous statistical methodology
  - We will use direct comparison to *in vivo* data in order to get an empirical estimate of our uncertainty
  - Any approximations, omissions, or mistakes should work to increase the estimated uncertainty when evaluated systematically across chemicals

# Statistical Analysis of High Throughput Toxicokinetics

United States Environmental Protection Agency



web2.elsevierproofcentra | web2.elsevierproofcentra | CRAN - Package httk

Secure | https://cran.r-project.org/web/packages/httk/index.html

Apps | DSStox | Confluence | JESEE | EHP | Battelle Box

**httk: High-Throughput Toxicokinetics**

Functions and data tables for simulation and statistical analysis of chemical toxicokinetics ("TK") using data obtained from relatively high throughput, in vitro studies. Both physiologically-based ("PBTK") and empirical (e.g., one compartment) "TK" models can be parameterized for several hundred chemicals and multiple species. These models are solved efficiently, often using compiled (C-based) code. A Monte Carlo sampler is included for simulating biological variability and measurement limitations. Functions are also provided for exporting "PBTK" models to "SBML" and "JARNAC" for use with other simulation software. These functions and data provide a set of tools for in vitro-in vivo extrapolation ("IVIVE") of high throughput screening data (e.g., ToxCast) to real-world exposures via reverse dosimetry (also known as "RTK").

| | |
|---|---|
| Version: | 1.6 |
| Depends: | R (≥ 2.10) |
| Imports: | deSolve, msm, data.table, survey, mvtnorm, truncnorm, stats, utils |
| Suggests: | ggplot2, knitr, rmarkdown, R.rsp, GGally, gplots, scales, EnvStats, MASS, RColorBrewer, TeachingDemos, classInt, ks, reshape2, gdata, viridis, CensRegMod, gmodels, colorspace |
| Published: | 2017-06-08 |
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| Maintainer: | John Wambaugh <wambaugh.john at epa.gov> |
| License: | GPL-3 |
| NeedsCompilation: | yes |
| Materials: | NEWS |
| CRAN checks: | httk results |

Downloads:

| | |
|---|---|
| Reference manual: | httk.pdf |
| Vignettes: | Creating Partition Coefficient Evaluation Plots<br>Age distributions<br>Global sensitivity analysis<br>Global sensitivity analysis plotting<br>Height and weight spline fits and residuals<br>Hematocrit spline fits and residuals<br>Plotting Css95<br>Serum creatinine spline fits and residuals<br>Generating subpopulations<br>Evaluating HTTK models for subpopulations<br>Generating Figure 2<br>Generating Figure 3<br>Plotting Howgate/Johnson data<br>AER plotting<br>Virtual study populations<br>httk: R Package for High-Throughput Toxicokinetics |

https://CRAN.R-project.org/package=httk

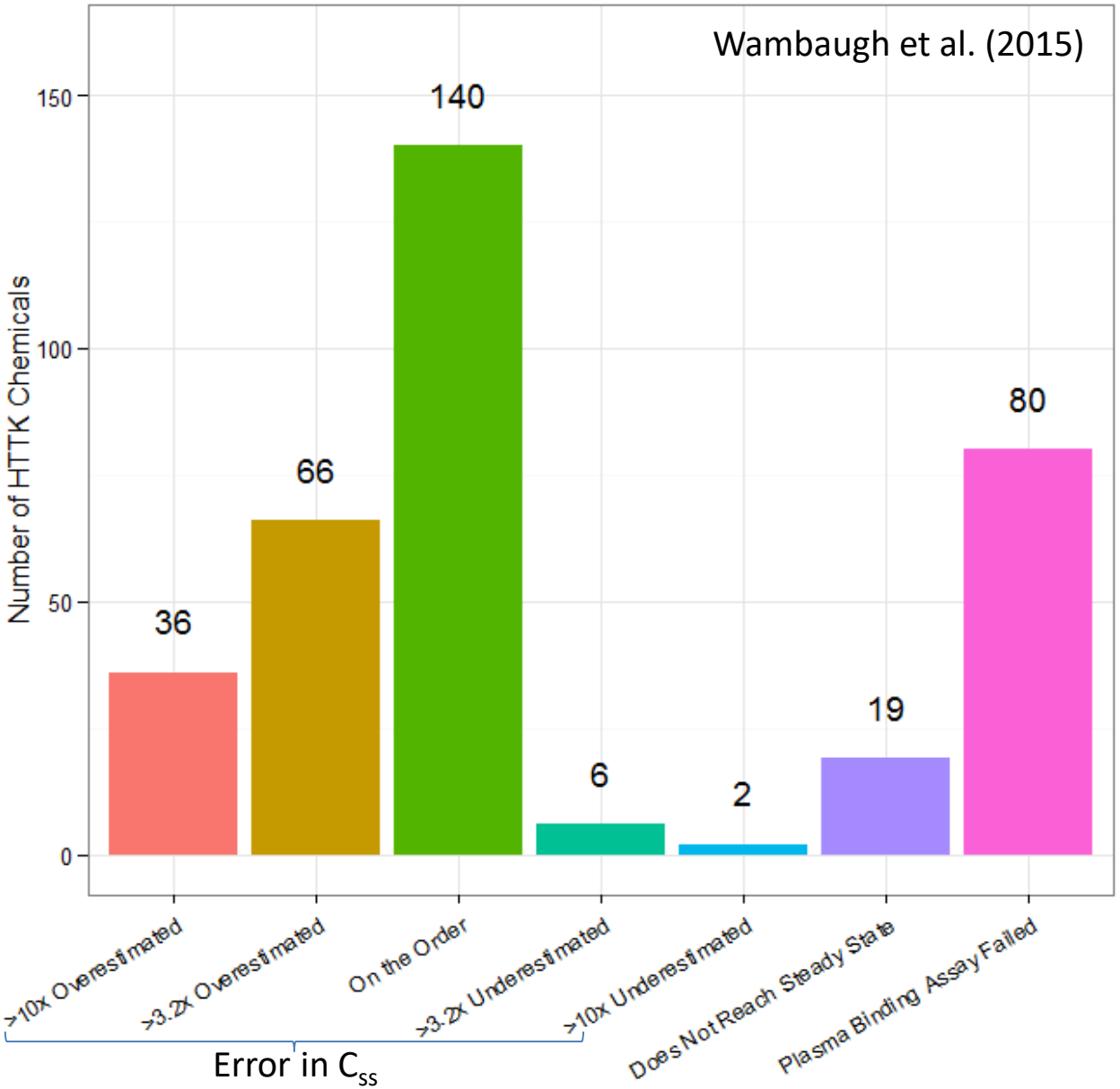
Can access this from the R GUI:

"Packages" then "Install Packages"

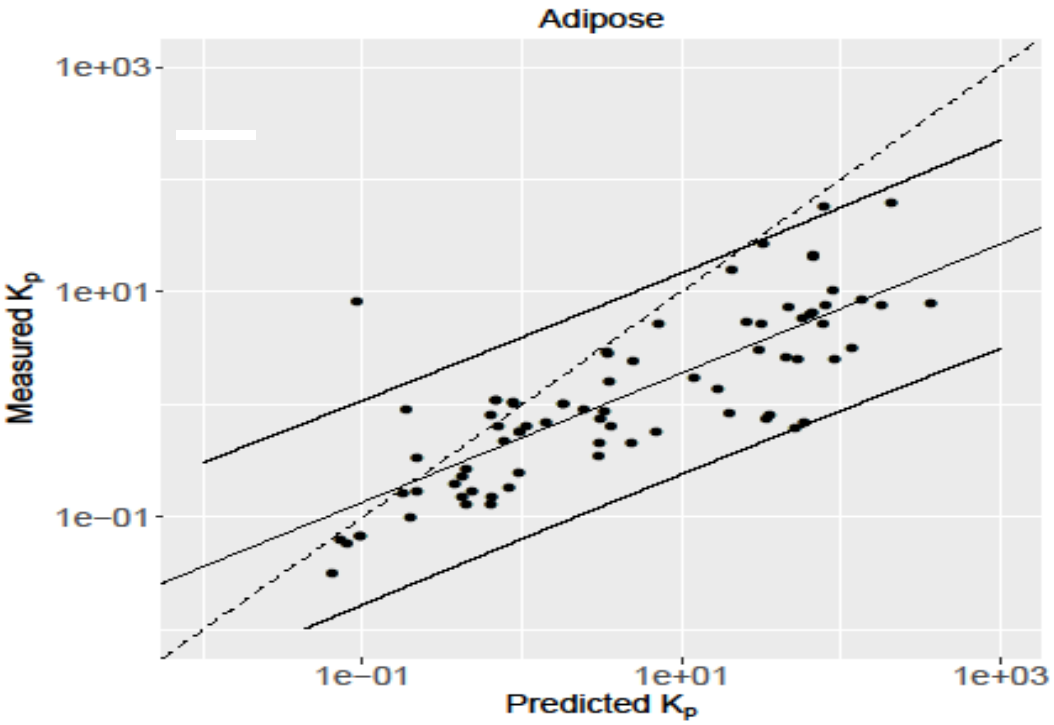
- "httk" R Package for *in vitro-in vivo* extrapolation and PBTK
- 553 chemicals to date
- 100's of additional chemicals being studied
- Pearce *et al.* (2017) provides documentation and examples
- Built-in vignettes provide further examples of how to use many functions

# Toxicokinetic Triage: Predicting TK IVIVE Errors

- Through comparison t*in vivo* data, a cross-validated (random forest) predictor of success or failure of HTTK has been constructed
- Add categories for chemicals that do not reach steady-state or for which plasma binding assay fails
- All chemicals can be placed into one of seven confidence categories



# Analyzing Legacy *In Vivo* Data




- Analyzed literature measurements of chemical-specific partition coefficients (PC) in rat
  - 945 tissue-specific PC
  - 137 unique chemicals
- Calibrating *in silico* predictors (Schmitt, 2008) to actual performance
  - Tissue-specific estimates of predictor bias and uncertainty
- Partition coefficient calibrations were evaluated with human measured volumes of distribution for 498 chemicals from Obach (2008)
  - Calibration to *in vivo* rat data improved predicted volume of distribution by a factor 3 for 116 chemicals



Pearce et al., (submitted)

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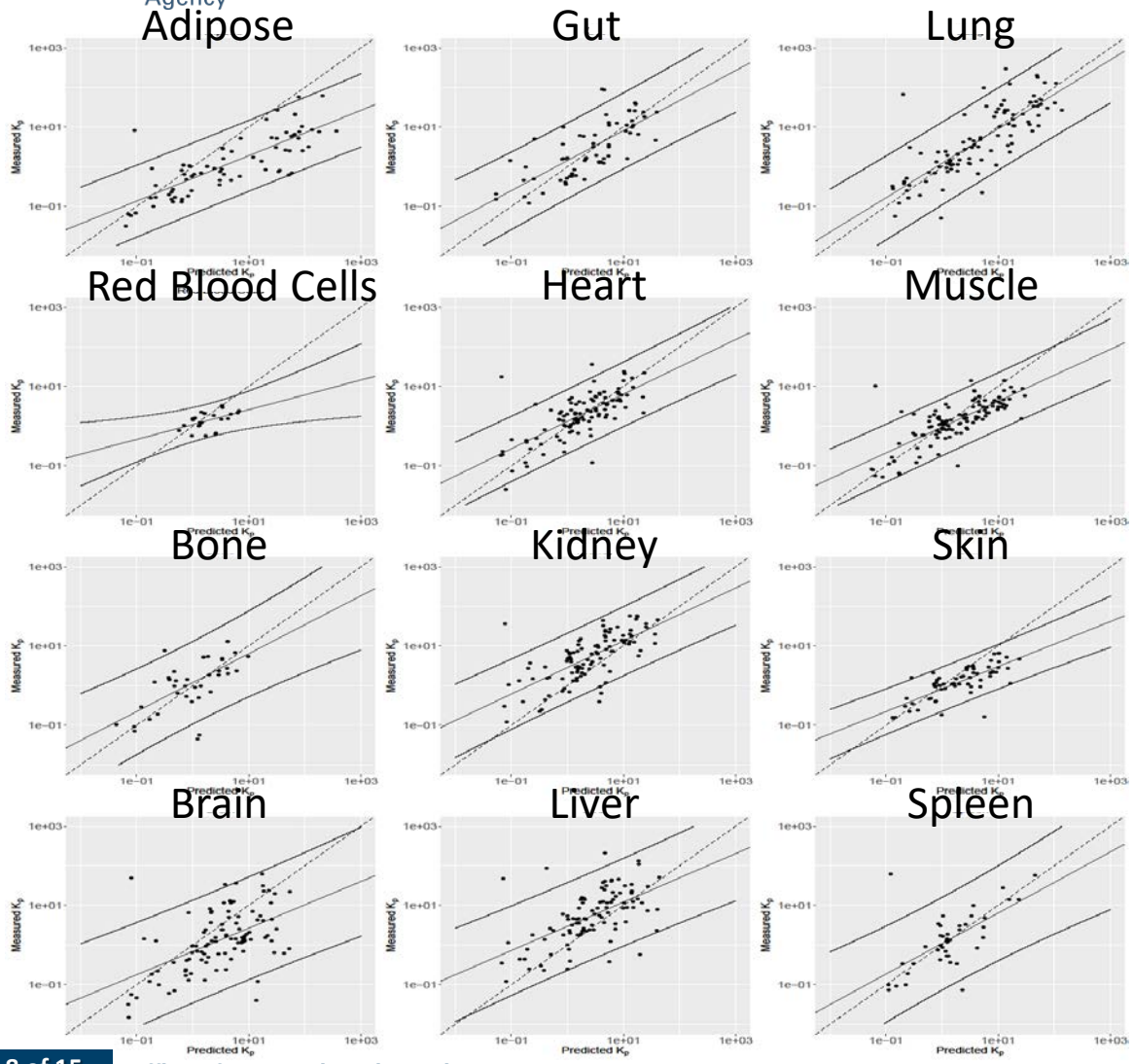


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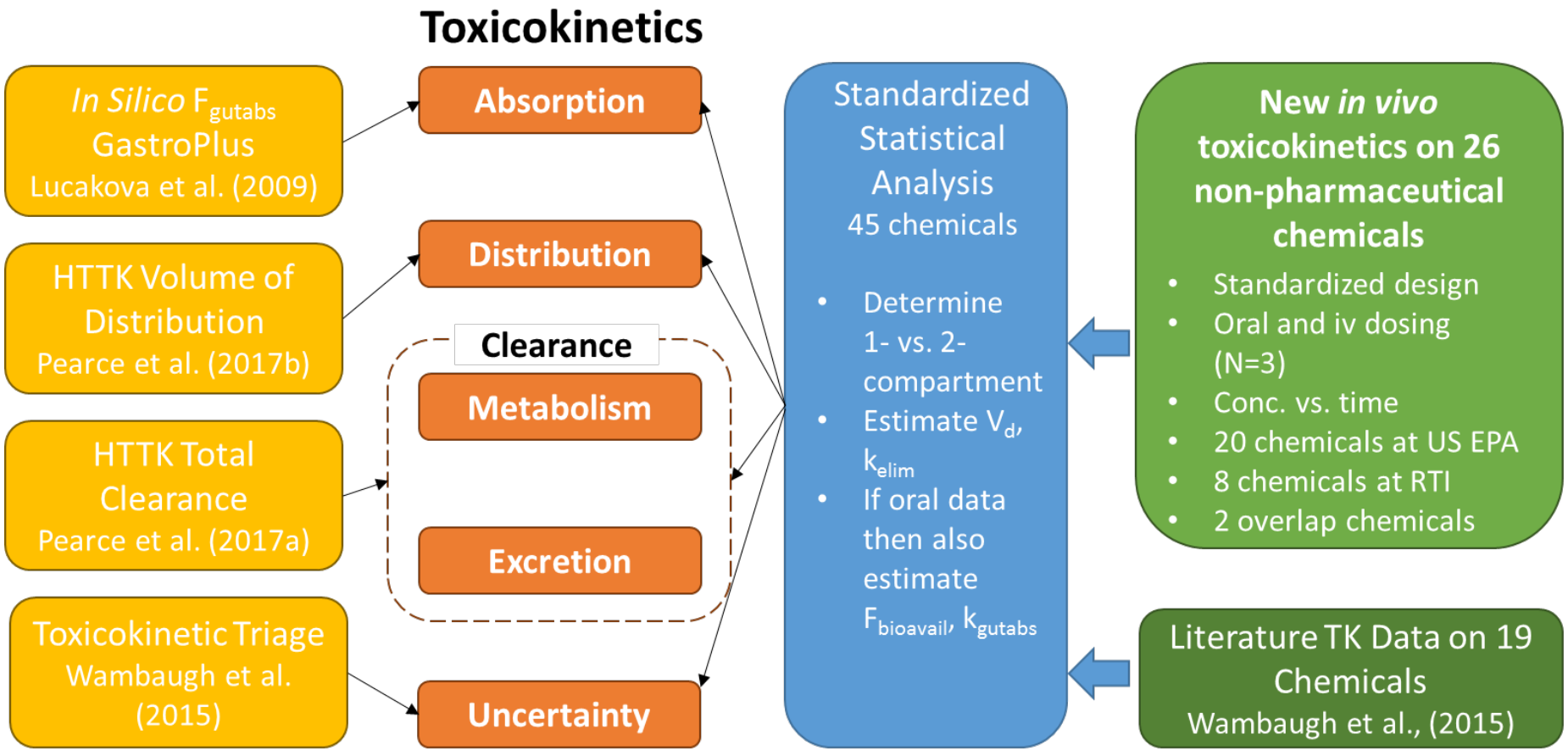
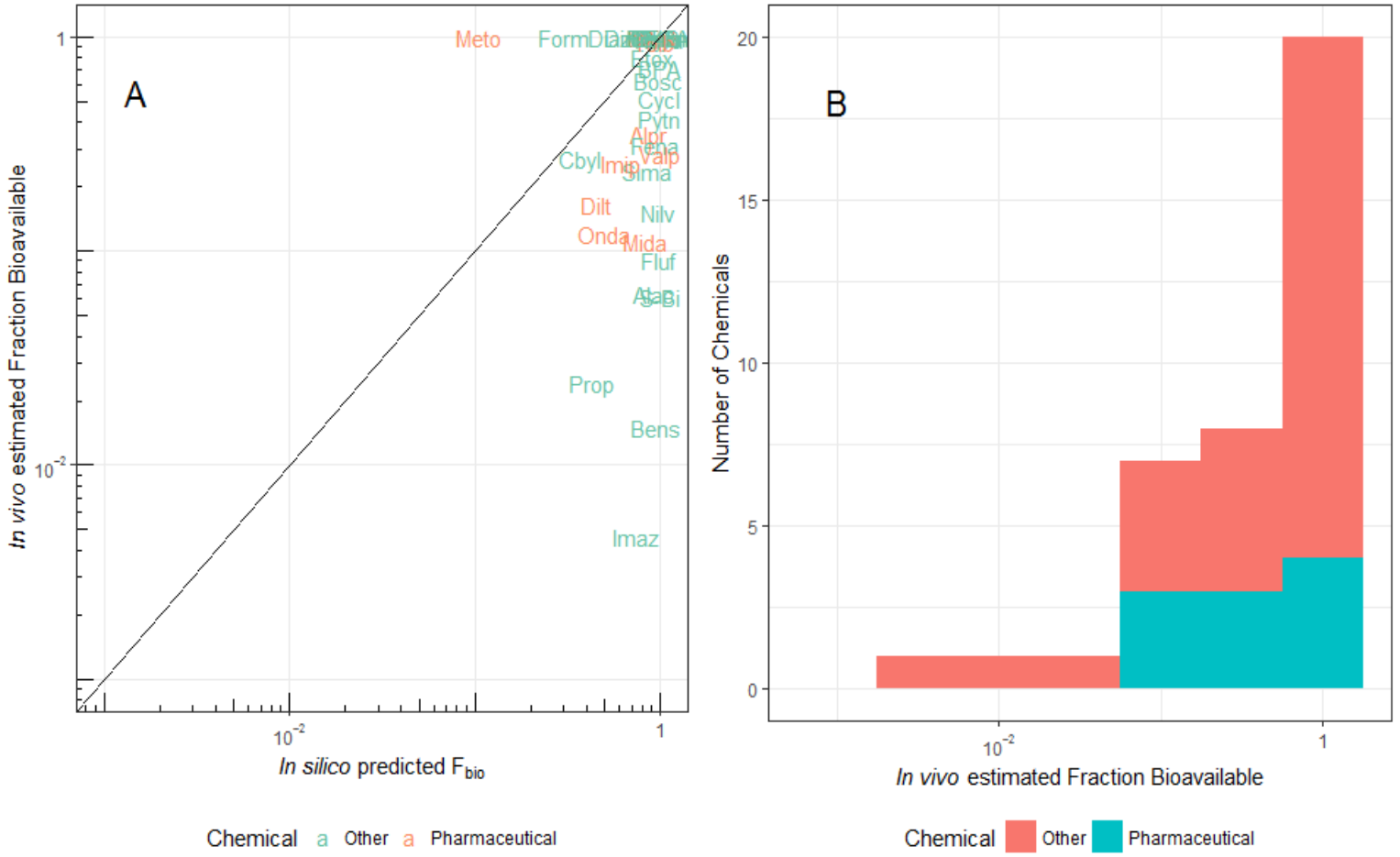
# Evaluating HTTK Predictions

26 chemicals more commonly associated with non-therapeutic and/or unintentional exposure



# Analyzing New *In Vivo* Data: Oral Absorption

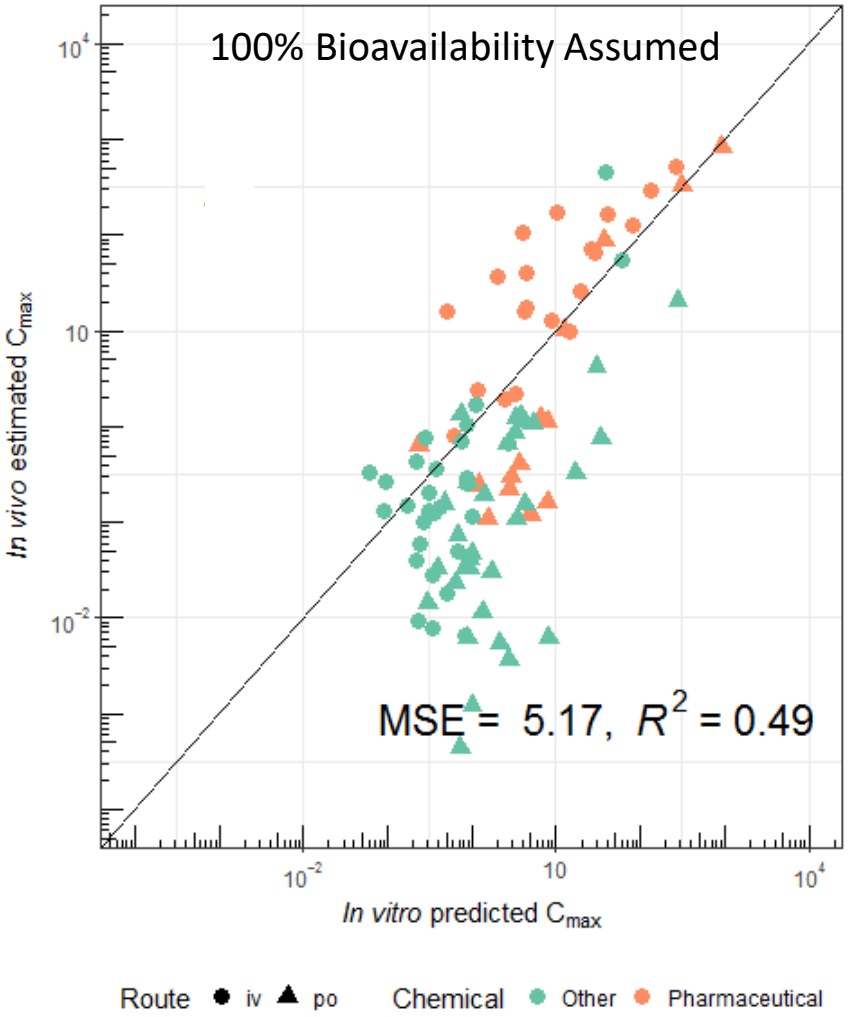




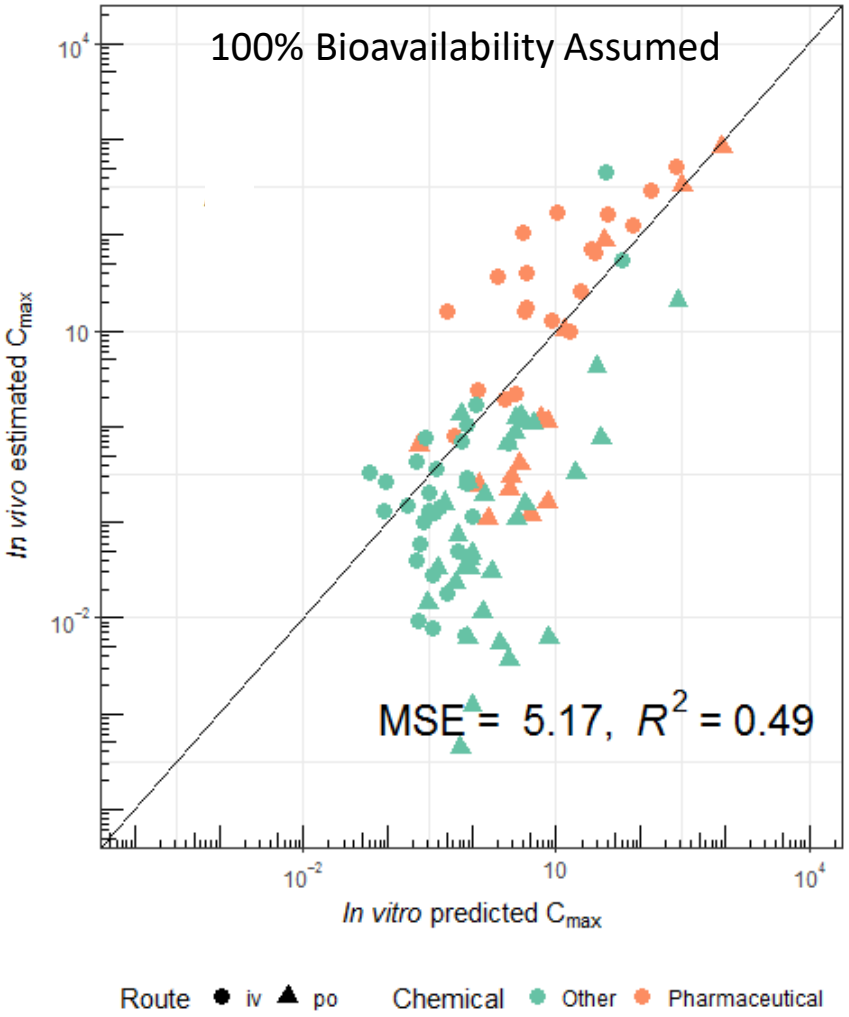

Bioavailability predictions from GastroPlus using only *in silico* values as input parameters (Nisha Sipes)

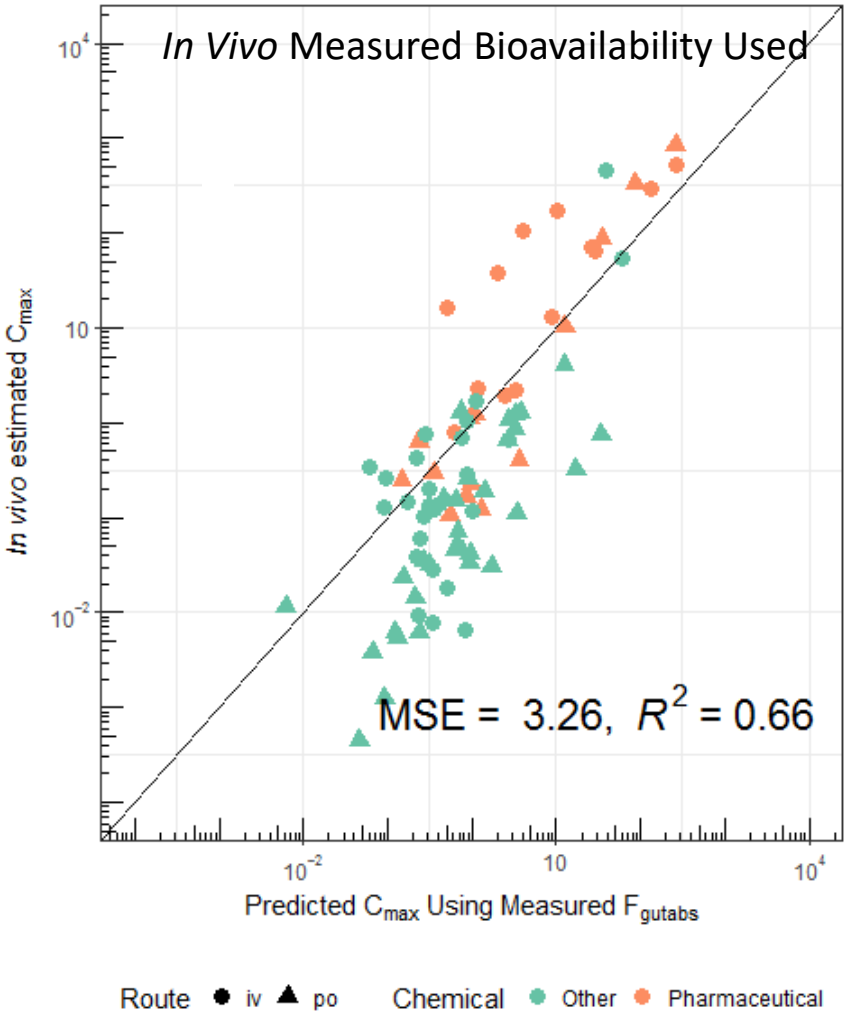
# Analyzing New *In Vivo* Data

United States Environmental Protection Agency



# Analyzing New *In Vivo* Data




**Cyprotex/Evotech (ToxCast) have now measured bioavailability (CACO2) for many HTTK chemicals**

# Can IVIVE + HTS Predict *In Vivo*?

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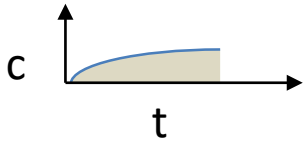
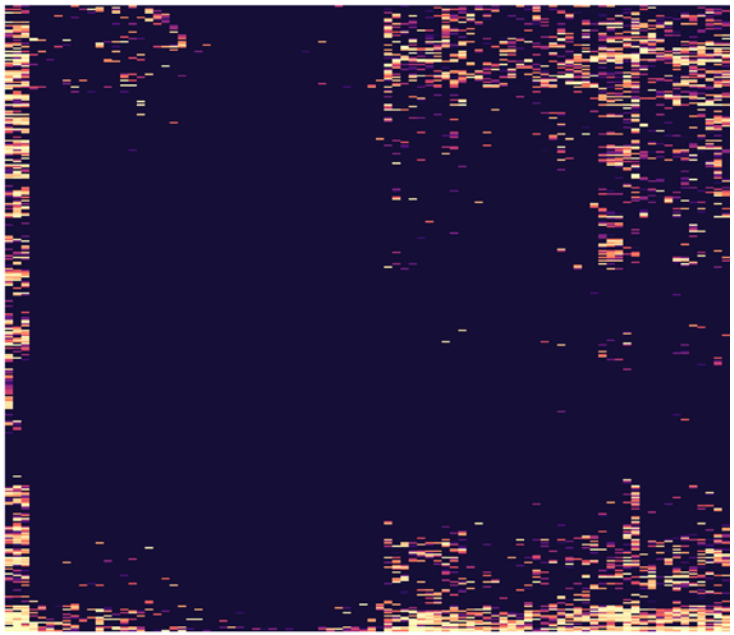
**ToxRefDB *in vivo* LEL dose (mg/kg/day)** → **HTTK** → **HTTK transformed concentration (µM)** **vs.** **ToxCast AC50 (µM)**




Plasma concentration determined by **HT-PBTK** shows **greater correlation** with **ToxCast AC50** than dose alone or y-randomization result

## HTTK *p*-Values




**HTTK** **Y-Randomized**



Analysis led by Greg Honda

# Exposure-Based Screening and Priority Setting

- R package "httk" freely available on CRAN allows statistical analyses of HTTK*
  1. We have **R**eused existing toxicokinetic (TK) data by compiling a library of TK time course data
     - Need non-pharma data: TK database being developed by Chris Grulke, Risa Sayre, Cecilia Tan
  2. Guided by IVIVE needs, we have **R**efined the design of *in vivo* TK studies,
  3. We use a **R**educed (n=6) study design including 3 intravenous and 3 orally dosed animals and tail blood sampling
  4. In some cases, we may be able to **R**eplace *in vivo* animal studies with HTS and HTTK
- HTTK methods appear to work for quantities like area under the curve (AUC), max plasma concentration ($C_{max}$) and steady-state serum concentration ($C_{ss}$)
- Bioavailability is a confounder – non-pharmaceuticals not well predicted by current tools
- **We can predict the doses at which some *in vivo* effects occur using *in vitro* data**



*Note that the open-source, free statistical language R is technically a fifth R here

EPA United States Environmental Protection Agency

# Chemical Safety for Sustainability (CSS) Rapid Exposure and Dosimetry (RED) Project

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**John Wambaugh**
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